Analysis of Cigarette Mainstream Smoke for 1,1-Dimethylhydrazine and Vinyl Acetate by Gas Chromatography–Mass Spectrometry

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Abstract

1,1-Dimethylhydrazine, also known as unsymdimethylhydrazine (UDMH) and vinyl acetate (VA), are both classified by the International Agency for Research on Cancer as 2B carcinogens (possibly carcinogenic to humans) and listed as cigarette smoke constituents; however, there is little or no quantitative data available on them. For UDMH in cigarette smoke, neither a yield nor a method has been published. For VA, the most recent information on yields dates back to 1965. To bridge this gap, we have developed new gas chromatographic–mass spectrometric methods for both compounds to determine their yields in cigarette smoke. UDMH is determined by derivatization with 2-nitrobenzaldehyde in methanol and is not found in cigarette smoke at levels above the detection limit of 19 ng/cig. In further experiments, when UDMH is added to the smoke stream or air stream of lit or unlit cigarettes, the derivative 2-nitrobenzaldehyde-2,2-dimethylhydrazone is found only in the air stream of the unlit cigarettes. From this, we conclude that UDMH is either not formed during smoking at all or, if it is, it reacts immediately and quantitatively with other smoke constituents (e.g., aldehydes) and is therefore not detectable in cigarette smoke. VA is determined by trapping in acetone at –78°C and is found at a concentration of 270 ng/cig for a standard reference cigarette with a cellulose acetate filter (the reference cigarette 1R4F). In the literature, VA is reported at concentrations of 1.6 µg/cig for a cigarette with a cellulose acetate/charcoal filter and 4 µg/cig for a cigarette with a cellulose acetate filter and for an unfiltered cigarette.

Introduction

1,1-Dimethylhydrazine, also known as unsymdimethylhydrazine (UDMH) and vinyl acetate (VA), are both classified by the International Agency for Research on Cancer (IARC) as 2B carcinogens (possibly carcinogenic to humans) and listed as cigarette smoke constituents (1–4).

UDMH is used in the synthesis of polymers, pesticides, pharmaceuticals, and chemotherapeutic agents (5,6). It is, together with monomethylhydrazine and hydrazine, a widely used rocket fuel (7). The toxicity of UDMH has been described in several publications (8–11) and is classified by IARC as a 2B carcinogen (possibly carcinogenic to humans) (1). Studies on laboratory animals have demonstrated the carcinogenicity and tumor-promoting activities of UDMH (12–14); however, very little information exists in the literature about UDMH in cigarette smoke, and the information available is unclear. There are two very similar publications from Hoffmann et al. (2,3) that report the occurrence of UDMH in cigarette smoke, but there is no reference for the data, no method, and no yield given. Another overview (4) of group 2B carcinogens in cigarette mainstream smoke does not list UDMH. One publication describes the occurrence of UDMH in unburned processed tobacco in which it was found in cigarette-blend, chewing tobacco, snuff, and Bright tobacco, but not in Burley tobacco (15). The U.S. Department of Health and Human Services suggests that if UDMH is found in tobacco, then smokers may be exposed to it (16). The source for UDMH in tobacco found by Schmeltz et al. (15) was quite probably the hydrolysis of the pesticide succinic acid 2,2-dimethylhydrazide (daminozide) (17). Daminozide is primarily used as a plant growth regulator on apples (22) and there is no information available citing its extensive use in tobacco plants. Different analytical techniques are reported in the literature for the determination of alkylated hydrazines. Because of the high reactivity of UDMH, all gas chromatographic (GC) methods require derivatization. GC analysis of underivatized UDMH will probably result in its adsorption or decomposition on the column, problems with sample stability in the injection port, broad peaks, and short column life. An intense background of many GC–mass spectrometry (MS) applications to mainstream cigarette smoke in the range of *m/z* 40 to 100 may also prevent the direct determination of UDMH. Several derivatization reagents have been used: pentafluorobenzaldehyde (15), pentafluorobenzoyl chloride (18), ace-

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tone (19,20), salicylaldehyde (21), and 2- and 4-nitrobenzaldehyde (22–25). We selected the 2-nitrobenzaldehyde derivatization method because it is easy to perform as well as fast, highly sensitive, selective, and quantitative. Saxton et al. (22), Majumdar et al. (23), and Brinkmann et al. (24) determined UDMH as a degradation product of daminozide. Inverse derivatizations are also cited in the literature. McDaniel and Howard (26) derivatized aldehydes, ketones, and carboxylic acids with UDMH, and VandenHeuvel and Horning (27) derivatized steroid ketones with UDMH. Because of the data reported by Hoffmann et al. (2,3), UDMH may be important when cigarette smoke is evaluated for its toxicological impact; therefore, a method was needed to be developed for the determination of UDMH in cigarette smoke.

The starting point for the method development for VA determination was the following: as opposed to its polymer, the monomer VA was used in the commercial production of polymers such as polyvinyl acetate (PVA) and copolymers such as ethylene-vinyl acetate in wood, paper, paint, and packaging and in the tobacco industry (28) as, for example, sideseam adhesives in the cigarette paper. Therefore, we evaluated the possibility that VA may be produced during combustion. The toxicity of VA has been described by Bogdanffy et al. (29) and classified by IARC as a 2B carcinogen (1,4). Studies on laboratory animals have demonstrated the carcinogenicity and tumor-promoting activities of VA (29,30). In 1965, Newsome et al. (31) detected VA in cigarette smoke at concentrations from 0.5 µg/puff (for a cigarette with a cellulose acetate filter and for an unfiltered cigarette) to 0.2 µg/puff (for a cigarette with a cellulose acetate/charcoal filter). Assuming that a cigarette has approximately 8 puffs, the values are approximately 4 µg/cig (unfiltered) and 1.6 µg/cig (charcoal filtered). It must be mentioned that the cigarette smoke was not generated under International Organization for Standardization (ISO) standard conditions and that the method used for quantitation was a GC–flame ionization detection (FID) method that, compared with the chromatographic possibilities of today, shows poor separation. Norman (32) reported a value of 4 µg/cig for VA, giving no reference for the data and no method for the determination of VA but citing Newsome et al. (31), one of his earlier coworkers. Guerin (33) reported 400 ng/cig citing the Norman article (32). This may have been a printer's error or a citation error; however, both values were cited in IARC (34). The overview of class 2B carcinogens in cigarette smoke by Smith et al. (4) also cited the Newsome et al. article (31). Headspace sampling (HS) techniques are mentioned in several publications for the determination of volatile VA. For example, Wang et al. (35) detected VA in coffee, and Kunigami et al. (36) described the analysis of VA in adhesive formulations. One publication from Watson and Ashley (37) analyzed acetates, including VA, in cigarette tobacco by HS–solid-phase microextraction, but no VA was detected (detection limit of 1.1 ng/cig). Because the most recent method for the determination of VA in cigarette smoke was a GC–FID method published in 1965 by Newsome et al. (31) and no more recent data were found in the literature, we decided to develop a method for the determination of VA in cigarette smoke.

Experimental

Materials

The test cigarettes were the University of Kentucky (Lexington, KY) reference cigarette 1R4F. This reference cigarette is one of a series of reference cigarettes developed for research purposes in a joint effort by the National Cancer Institute of the National Institute of Health (Bethesda, MD), the Agriculture Research Service of the U.S. Department of Agriculture (USDA) (Washington, D.C.), and the University of Kentucky Tobacco and Health Research Institute (Lexington, KY). The reference cigarette 1R4F was designed in 1983 with the following specifications: 83.5-mm length, 25-mm circumference, 35-mm butt length, 11-mg total particular matter (TPM), and 0.8-mg nicotine/cig. The cigarettes were stored and conditioned according to ISO standard 3402 (38).

Reagents and chemicals

UDMH, 2-nitrobenzaldehyde, VA, acetaldehyde, and propionaldehyde were purchased from Aldrich, Fluka, and Sigma (Deisenhofen, Germany) and were labeled as 98% or 99% pure. VA-d_6 (99% pure) was obtained from CDN Isotopes (Dr. Ehrenstorfer, Augsburg, Germany). Methanol (Lichrosolv) and acetone (p.A.) were purchased from Merck (Darmstadt, Germany).

Preparation of standards and derivatization reagent

A stock solution of UDMH was prepared in methanol at a concentration of 1.55 mg/mL. From this stock solution, standard solutions were prepared for external calibration and to verify the linearity of the derivatization. Acetaldehyde and propionaldehyde were prepared in methanol at concentrations of 20 µL/mL or were used as pure solutions. The derivatization reagent was prepared with 2-nitrobenzaldehyde dissolved in methanol at concentrations of approximately 100 mg/mL.

Stock solutions of VA and VA-d_6 were prepared in acetone at concentrations of 8.59 and 7.04 mg/mL. From these stock solutions, standard solutions were prepared for an internal standard calibration. A stock solution of 4.1 mg/mL was prepared for control samples of VA.

Smoke generation, sample collection, and derivatization of UDMH

Mainstream smoke was generated in basic conformity with ISO 3308 (39). The cigarettes were electrically lighted and smoked on a 20-port rotary smoking machine with a 35- ± 0.5-mL puff volume and a 2.0- ± 0.1-s puff duration every minute. The cigarette mainstream smoke was collected under conditions adapted from a published method (40). UDMH was collected in only one (the first) wash bottle (midget microimpinger), whereas VA was collected in 3 wash bottles connected in series (Figure 1). The first wash bottle contained approximately 6 g of glass beads, and the second and the third wash bottles each contained approximately 5 g of glass beads. For the determination of UDMH, the first wash bottle contained 5 mL methanol and 2 mL derivatization reagent, and no glass fiber filter was used. For the determination of VA, the first wash bottle contained 7 mL acetone, and the second and third

wash bottles each contained approximately 6 mL acetone. Prior to sampling, the wash bottles were cooled to –78°C with a mixture of isopropanol and dry ice. After sampling was completed, the wash bottles were thawed and the contents funneled into a volumetric flask. The wash bottles were rinsed with solvent and the volumetric flask was filled to the 10-mL mark for UDMH and the 25-mL mark for VA. One milliliter of the sample was added to an autosampler vial. The UDMH sample was heated for 30 min at 30°C. The internal standard VA-d_6 was added to the VA sample.

In some experiments for the determination of UDMH, the cigarettes were not lit ("dry puffs"); instead, fresh air was drawn through the cigarettes, and the sample collection was performed in the same way as with the lit cigarettes.

Standard addition experiments for the determination of UDMH

UDMH was added to the smoke stream through a glass connector placed between the smoking machine and the wash bottles (Figure 1). After the second, fourth, fifth, and seventh puffs of the smoking machine, 10 µL of a standard solution of UDMH containing 1.55 mg/mL was added to the smoke (total 62 µg UDMH). After each addition, the hole was closed. In some experiments, a hairdryer was used to heat the glass connector to insure that all the added UDMH (boiling point 63.9°C) vaporized. In a separate experiment, 5 µL of a standard solution of UDMH (concentration of 1.55 mg/mL) was added to cigarette filters (7.7 µg/filter). The cigarettes were then smoked, and after smoking, the filters were cleaned of tobacco. The filters were extracted with a solution of 8 mL methanol and 2 mL derivatization reagent by shaking for 10 min. An aliquot of the solution was removed and heated for 30 min at 30°C for derivatization.

GC–MS instrumentation

A Hewlett Packard (HP) 6890 GC equipped with an HP 6890 Series autosampler was coupled with an HP5973 mass-selective detector (230°C transfer line temperature) operating in electron impact (70 eV) mode. The MS quadrupole and source heaters were maintained at 106°C and 200°C, respectively. The GC worked in splitless mode and was fitted with an SGE BP 624 column with a 0.32-mm i.d. and 1.8-µm film thickness. Helium carrier gas was maintained at 2.6 mL/min. For the determination of 2-nitrobenzaldehyde–2,2-dimethylhydrazone (2-NDH), the GC oven was heated to 100°C and held for 2 min, then increased to 230°C at 30°C/min, and held for 6 min (total GC run time 12.33 min). For the determination of acetaldehyde 1,1-dimethylhydrazone and propionaldehyde 1,1-dimethylhydrazone, the GC oven was held at 50°C for 2 min, then increased to 230°C at 10°C/min, and held for 3 min (total GC run time 23 min). For the determination of VA, the GC oven was started at 40°C, held for 3 min and heated at 50°C to 200°C, and then held for 1 min (total GC run time 7.20 min).

MS parameters and data analysis

Mass spectra from standard solutions of the hydrazones were acquired in full-scan mode (mass range of 50 to 400). For the quantitation of 2-NDH, selected masses were acquired in single-ion monitoring (SIM) mode; 2-NDH was quantitated by its molecular ion $(M)^+$ *m/z* 193 and the ion at *m/z* 74 was used for confirmation. An additional criterion for the determination of 2-NDH was the retention time of 9.5 min (±0.05 min). Acetaldehyde 1,1-dimethylhydrazone was detected with the $(M)^+$ ion at *m/z* 86 and propionaldehyde 1,1-dimethylhydrazone with the $(M)^+$ ion at *m/z* 100 in SIM mode.

VA and VA-d_6 were detected and quantitated in SIM mode with the masses of the molecular ions (*m/z* 86 and 92). Qualifier ions for confirmation could not be used because of matrix effects.

For data analysis, the chromatogram peak areas were determined automatically by the HP ChemStation Integrator program in the HP Enhanced ChemStation Version B.03.00 software.

Calibration and validation data

Calibration and validation data for the determination of UDMH

A 12-point calibration curve for 2-NDH with a concentration range of 0.077 to 15.47 µg/mL was produced as an external calibration and to verify the linearity of the derivatization. The calibration curve was linear (correlation coefficient $[r^2] = 0.999$) and all of the results were within 8% of expected. The concentration of 2-NDH at 0.08 µg/mL was found to be the quantitation limit with a signal-to-noise (S/N) ratio of 10:1, and the concentration of 0.03 µg/mL was found to be the detection limit (S/N = 3:1). Assuming that 10 cigarettes would be smoked and the smoke trapped in a 7-mL derivatization reagent in methanol, the detection limit would be 19 ng/cig. The stability of 2-NDH in the matrix of 10 cigarettes smoked and trapped in methanol was

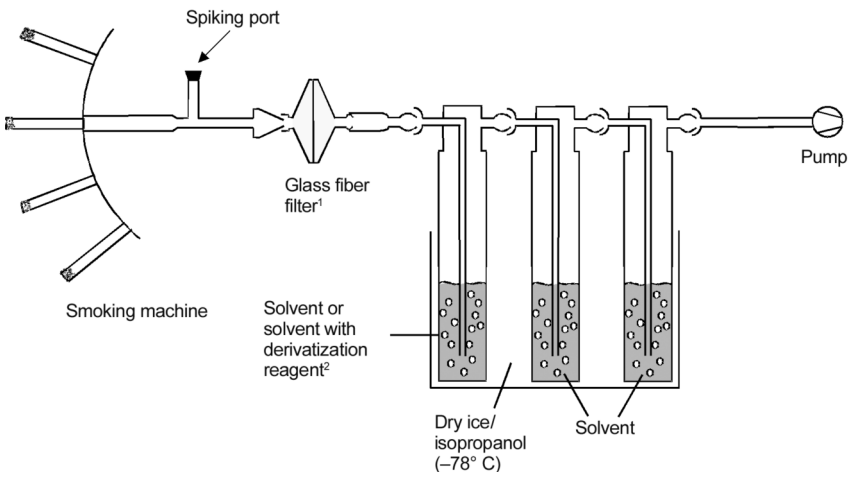

Figure 1. A collection of gas-phase cigarette mainstream smoke: (1) glass fiber filter was not used for the UDMH determination and (2) only the first wash bottle was used for the UDMH determination

determined by the following procedure: a standard solution of UDMH and 2-nitrobenzaldehyde was added to the microimpingers filled with methanol. One control sample was removed for measuring. Then, ten cigarettes were smoked, and the smoke was trapped in the microimpingers. Samples were taken out immediately and again 6 h after smoking. There was no loss of 2-NDH in either sample. In one sample, which was removed 72 h after sample trapping, there was a 28% decrease in 2-NDH. Based on these results, all experiments (sample collection, extraction, derivatization, and measurement) were carried out within 6 h. The decrease in concentration may have been the result of stability problems with hydrazones in the presence of light and an acetic acid catalyst (5,20).

Calibration and validation data for the determination of VA

An 18-point calibration curve for VA with a concentration range of 0.02 to 85.9 µg/mL was linear (r^2 = 1.0) and all of the results were within 2.2% of expected. The concentration of VA at 0.02 µg/mL was found to be the quantitation limit (S/N = 10:1). Assuming that 10 cigarettes would be smoked and the smoke trapped in 25 mL acetone, the quantitation limit would be 8 ng/cig. The precision of an intraday (n = 5) determination of samples from 10 1R4F cigarettes was 2.5%. The trapping efficiency was 82% in the first trap, 16% in the second trap, 2% in the third trap, and the concentration of VA in a fourth wash bottle, connected in series to the other three, was below the detection limit.

Results

UDMH in cigarette smoke from the reference cigarette 1R4F

Smoke was generated from 10 1R4F cigarettes, and the smoke constituents were trapped in a microimpinger at –78°C filled with methanol and the derivatization reagent. After derivatization, the liquid trap was analyzed for 2-NDH (Table I). No 2-NDH was found. In addition, approximately 62 µg UDMH was added to the smoke via the spiking port (Figure 1), but the derivative 2-NDH was still not found, even when only one cigarette was smoked.

In contrast, when UDMH was added to the "airstream" of unlit cigarettes, the derivative was found (Table I). We assume that UDMH does not reach the trap, because it reacts with smoke components (e.g., with aliphatic aldehydes or ketones). Bark and Prachuabpaibul (41) and McDaniel and Howard (26) reported that aliphatic aldehydes react quantitatively with UDMH, and Smith et al. (42) obtained good yields for the reaction of aliphatic steric unhindered ketones with UDMH.

Table I. Detection of 2-NDH in Liquid Traps Before and After UDMH was Added to the Smoke or Airstream of Lit or Unlit Cigarettes

Cigarettes	No. of cigarettes	UDMH added to smoke/airstream	2-NDH found above the detection limit (19 ng/cig)
Lit	10	no	no
Lit	10	yes	no
Lit	1	yes	no
Unlit	10	yes	yes

With regard to the possibility that UDMH may have remained on the cellulose acetate of the cigarette filter, we performed the following experiments: we added UDMH to the cigarette filters and "smoked" the cigarettes lit and unlit. The extracts from the filters showed that UDMH disappears in smoke from the lit cigarettes, whereas it could be extracted from the filter of the unlit cigarettes. We assumed that during smoking, any UDMH that may be formed reacts with other smoke constituents (e.g., aldehydes). To explore this hypothesis, we bubbled the cigarette mainstream constituents acetaldehyde and propionaldehyde through a methanolic solution of UDMH. Although it was easy to identify the corresponding acetaldehyde 1,1-dimethylhydrazone and propionaldehyde 1,1-dimethylhydrazone by their mass spectra, attempts to analyze cigarette smoke extract for the hydrazones were not successful.

Concentration of VA in the cigarette smoke from the reference cigarette 1R4F

Smoke was generated from 10 1R4F cigarettes, and the smoke constituents were trapped in 3 microimpingers connected in series at –78°C filled with acetone. A concentration of 270 ng/cig was found.

Conclusion

We have developed reliable methods for the determination of UDMH and VA in cigarette smoke and found that UDMH is not detectable in cigarette smoke at levels above 19 ng/cig with the analytical method described. Through standard addition experiments, we found that UDMH is not stable in cigarette smoke, which means that if UDMH is initially formed during smoking, its reaction with other smoke constituents is immediate and quantitative. It is worth noting that UDMH may be a precursor of *N*-nitrosodimethylamine (NDMA), a class 2A carcinogen that occurs in cigarette smoke up to 1.62 µg/cig (43), and that oxidative processes on UDMH can tend to produce NDMA (44,45); however, this has not been shown to take place in cigarette smoke.

The VA yield in cigarette smoke was 270 ng/cig for the reference cigarette 1R4F. The method can be used, for example, to investigate the influence of PVA-based adhesives used in cigarette production on cigarette smoke composition.

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References

1. IARC. Overall evaluations of carcinogenicity: an updating of IARC monographs volumes 1–42. *IARC Monographs Suppl.* **7:** 37–74 (1987).
2. D. Hoffmann, I. Hoffmann, and E.L. Wynder. *The Changing Cigarette,* 1950–1997: Facts and Expectations. Proc. Canada Conference of Cigarette Toxicity Reduction. W.S. Rickert, Ed. Toronto, Ontario, Canada, 1998, pp. 1–94.
3. D. Hoffmann, I. Hoffmann, and K. El-Bayoumy. The less harmful cigarette: a controversial issue. A tribute to Ernst L. Wynder. *Chem. Res. Toxicol.* **14:** 767–90 (2001).
4. C.J. Smith, T.A. Perfetti, A. Rumple, A. Rodgman, and D.J. Doolittle. "IARC group 2B carcinogens" reported in cigarette mainstream smoke. *Food Chem. Toxicol.* **39:** 183–205 (2001).
5. S. Cathum, V. Atamaniouk, L. Ananieva, C. Ladanowski, and H. Whittaker. Gas chromatography–mass spectrometric determination of unsymdimethyl-hydrazine in soil and water by derivatization with aromatic aldehydes. *Can. J. Chem. Engin.* **76:** 680–85 (1998).
6. V.A. Lopyrev, G.V. Dolgushin, and M.G. Voronkov. Applied chemistry of 1,1-dimethylhydrazine and its derivatives. *Russ. J. Appl. Chem.* **71:** 1295–1309 (1998).
7. L.A. Fedorov. Liquid missile propellants in the former Soviet Union. *Environ. Pollut.* **105:** 157–61 (1999).
8. J.P. Rouganne, A. Cierand, and P. Duca. Effets hépatiques d'une intoxication subaiquë par l'hydrazine et la diméthyl-hydrazine chez la souris. *C.R. Soc. Biol.* **163:** 192–95 (1962).
9. C.F. Simpson and M.V. Barrow. Toxicity of a substituted hydrazine for turkeys. *Arch. Environ. Health* **25:** 349–53 (1972).
10. J.-P. Chevrier and A. Pfister. Étude sur la toxicité de la 1,1 dimethylhydrazine chez l'animal: 2. Intoxication chronique. *Eur. J. Toxicol. Environ. Hyg.* **7:** 242–46 (1974).
11. M.L. Barth, C.L. Geakeland, and H.H. Cornish. 1,1-Dimethylhydrazine induced diuresis. *Toxicol. Appl. Pharmacol.* **11:** 26–34 (1967).
12. M. Sakita, H. Imai, M. Kasuga, N. Kageyama, S. Imaki, M. Tamai, Y. Fujita, and S. Majima. Effect of a protein-bound polysaccharide preparation, PS-K, on dimethyl hydrazine induction of intestinal tumors in rats. *Gann.* **74:** 351–59 (1983).
13. O. Kimura, N. Kaibara, Y. Miyano, T. Okamoto, H. Tamura, E. Yurugi, and S. Koga. Nuclear DNA content in dimethylhydrazine-induced colonic carcinoma and muscosal dysplasia in rats. *Cancer* **53:** 1918–22 (1984).
14. T. Tamura, M. Shibutani, K. Toyoda, T. Shoda, K. Takada, C. Uneyama, M. Takahashi, and M. Hirose. Tumor-promoting activities of hydroqinone and 1,1-dimethylhydrazine after initiation of newborn mice with 1-methyl-1-nitrosourea. *Cancer Lett.* **143:** 71–80 (1999).
15. I. Schmeltz, S. Abidi, and D. Hoffmann. Tumorigenic agents in unburned processed tobacco: N-nitrosodiethanolamine and 1,1-dimethylhydrazine. *Cancer Lett.* **2:** 125–32 (1977).
16. U.S. Department of Health and Human Services. National Toxicology Program. Report on Carcinogens, 9th ed. 2001. http://ehp.niehs.nih.gov/roc/ninth/rahc/1_1dimethylehydrazine.pdf. National Institute of Environmental Health Sciences, Triangle Park, NC.
17. D. Hoffmann. Personal communication (2002).
18. M.A. Rutschmann and H.-R. Buser. Determination of daminozide and dimethylhydrazine residues in Swiss apple juice concentrates using gas chromatography–mass spectrometry. *J. Agric. Food Chem.* **39:** 176–81 (1991).
19. J.T. Veal. The analysis of hydrazine, monomethyl hydrazine and 1,1-dimethylhydrazine using GC techniques. *Proc. Conf. Environ. Chem.* 79–97 (1977).
20. J.R. Holtzclaw, S.L. Rose, J.R. Wyatt, D.P. Rounbehler, and D.H. Fine. Simultanous determination of hydrazine, methylhydrazine, and 1,1-dimethylhydrazine in air by derivatization/gas chromatography. *Anal. Chem.* **56:** 2952–56 (1984).
21. K.T. Faughnan and M.A. Woodruff. Modified gas chromatographic/mass spectrometric method for the determination of daminozide in high protein food products. *J. Assoc. Off. Anal. Chem.* **74:** 682–92 (1991).
22. W.L. Saxton, K. Steinbrecher, and E. Gunderson. Results of a survey for the presence of daminozide and unsymmetrical dimethylhydrazine in food. *J. Agric. Food Chem.* **37:** 570–73 (1989).
23. T.K. Majumdar, P.W. Geno, and A. Yau. Determination of daminozide residues at very low levels in fruits by gas chromatography/mass spectrometry. *J. Agric. Food Chem.* **43:** 1421–23 (1995).
24. J.H. Brinkman, A.G. van Dijk, R. Wagenaar, and J.K. Quirijns. Determination of daminozide residues in apples using gas chromatography with nitrogen-phosphorous detection. *J. Chromatogr. A* **723:** 355–60 (1996).
25. D.P. Samsonov, G.V. Bornovalova, R.I. Pervunina, and N.P. Zhiryukhina. Determination of N,N-dimehtylhydrazine in soil by gas chromatography–mass spectrometry. *J. Anal. Chem.* **53:** 169–72 (1998).
26. C.A. McDaniel and R.W. Howard. Mass spectral determination of aldehydes, ketones, and carboxylic acids using 1,1-dimethylhydrazine. *J. Chem. Ecol.* **11:** 303–10 (1985).
27. W.J.A. VandenHeuvel and E.C Horning. Gas-chromatographic characterization of steroid ketones as N,N-dimethylhydrazones. *Biochim. Biophys. Acta* **74:** 560–63 (1963).
28. C.N. Kunigami, M.S. Sanctos, and M.H.W. Morelli-Cardoso. Development of a methodology for the identification of volatiles in industrial adhesive formulations by GC/MS with manual headspace sampling. *J. High Resol. Chromatogr.* **22(8):** 477–80 (1999).
29. M.S. Bogdanffy, T.R. Tyler, M.B. Vinegar, R.W. Rickard, F.M.B. Carpanini, and T.C. Cascieri. Chronic exposure and oncogenicity study with vinyl acetate in the rat. In Utero exposure in drinking water. *Fundam Appl. Toxicol.* **23:** 206–14 (1994).
30. M.S. Bogdanffy, D.R. Plowchalk, R. Sarangapani, T.B. Starr, and M.E. Andersen. Mode-of-action-based dosimeters for interspecies extrapolation of vinyl acetate inhalation risk. *Inhal. Toxicol.* **13:** 377–96 (2001).
31. J.R. Newsome, V. Norman, and C.H. Keith. Vapor phase analysis of tobacco smoke. *Tob. Sci.* **4:** 24–32 (1965).
32. V. Norman. An overview of the vapor phase, semivolatile and nonvolatile components of cigarette smoke. *Recent Adv. Tob. Sci.* **3:** 28–58 (1977).
33. M.R. Guerin. "Chemical Composition of Cigarette Smoke." In Banbury Report: A Safe Cigarette? G.B. Gori and F.G. Bock, Eds. CSH Press, Cold Spring Harbor, NY, 1980, Vol. 3, pp. 191–204.
34. IARC. Monographs on the evaluation of carcinogenic risks to human. *IARC Monographs Suppl.* **63:** 443–65 (1995).
35. T.H. Wang, H. Shanfield, and A. Zlatkis. Analysis of trace volatile organic compounds in coffee by headspace concentration and gas chromatography–mass spectrometry. *Chromatographia* **17:** 411–17 (1983).
36. C.N. Kunigami, M.S. Sanctos, and M.H.W. Morelli-Cardoso. Development of a methodology for the identification of volatiles in industrial adhesive formulations by GC/MS with manual headspace sampling. *J. High Resol. Chromatogr.* **22(8):** 477–80 (1999).
37. C.H. Watson and D.L. Ashley. Quantitative analysis of acetates in cigarette tobacco using solid-phase microextraction and gas chromatography–mass spectrometry. *J. Chromatogr. Sci.* **38:** 137–44 (2000).
38. International Standard ISO 3402, *Tobacco and Tobacco Products—Atmosphere for Conditioning and Testing,* 3rd ed. 1991. International Organization for Standardization, Genève, Switzerland.
39. International Standard ISO 3308, *Cigarettes—Routine Analytical Cigarette Smoking Machine—Definitions and Standard Conditions,* 3rd ed. 1991. International Organization for Standardization, Genève, Switzerland.
40. G.D. Byrd, K.W. Fowler, R.D. Hicks, M.E. Lovette, and M.F. Borgerding. Isotope dilution gas chromatography–mass spectrometry in the determination of benzene, toluene, styrene and

acrylonitrile in mainstream cigarette smoke. *J. Chromatogr.* **503:** 359–68 (1990).
41. L.S. Bark and P. Prachuabpaibul. The thermometric determination of some aldehydes by hydrazone formation with unsymmetrical dimethylhydrazine. *Anal. Chim. Acta* **84:** 207–10 (1976).
42. R.F. Smith, J.L. Marucci, and P.S. Tingue. Base-promoted elimination reactions of acetaldehyde N-alkyl-N,N-dimethylhydrazonium salts. A convient synthesis of N,N-dimethylalkylamines. *Synth. Commun.* **22:** 381–89 (1992).
43. C.J. Smith, T.A. Perfetti, M.A. Rumple, A. Rodgman, and D.J. Doolittle. "IARC group 2A carcinogens" reported in cigarette mainstream smoke. *Food Chem. Toxicol.* **38:** 371–83 (2000).
44. M. Castegnaro, I. Brouet, J. Michelon, G. Lunn, and E.B. Sansone. Oxidative destruction of hydrazines produces N-nitrosamines and other mutagenic species. *Am. Ind. Hyg. Assoc. J.* **47:** 360–64 (1986).
45. G. Lunn and E.B. Sansone. Oxidation of 1,1-dimethylhydrazine (UDMH) in aqueous solution with air and hydrogen peroxide. *Chemosphere* **29:** 1577–90 (1994).

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